Review

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Current understanding and future directions in the application of TIMP-2 and IGFBP7 in AKI clinical practice

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Abstract: NephroCheck® is the commercial name of a combined product of two urinary biomarkers, tissue inhibitor of metalloproteinases-2 (TIMP-2) and insulin-like growth factor-binding protein 7 (IGFBP7), expressed as [TIMP-2] · [IGFBP7], used to identify patients at high risk of acute kidney injury (AKI). AKI is a common and harmful complication especially in critically-ill patients, which can induce devastating short- and long-term outcomes. Over the past decade, numerous clinical studies have evaluated the utility of several biomarkers (e.g. neutrophil gelatinase-associated lipocalin, interleukin-18, liver-type fatty acid binding protein and kidney injury molecule-1, cystatin C) in the early diagnosis and risk stratification of AKI. Among all these biomarkers, [TIMP-2] · [IGFBP7] was confirmed to be superior in early detection of AKI, before the decrease of renal function is evident. In 2014, the US Food and Drug Administration permitted marketing

of NephroCheck® (Astute Medical) (measuring urinary [TIMP-2] · [IGFBP7]) to determine if certain critically-ill patients are at risk of developing moderate to severe AKI. It has since been applied to clinical work in many hospitals of the United States and Europe to improve the diagnostic accuracy and outcomes of AKI patients. Now, more and more research is devoted to the evaluation of its application value, meaning and method in different clinical settings. In this review, we summarize the current research status of [TIMP-2] · [IGFBP7] and point out its future directions.

Keywords: acute kidney injury (AKI); biomarker; cell cycle arrest; insulin-like growth factor-binding protein 7 (IGFBP7); NephroCheck®; tissue inhibitor of metalloproteinases-2 (TIMP-2).

Introduction

Acute kidney injury (AKI) is a multifactorial disease. It commonly complicates high-risk surgeries and appears as a consequence of systemic illness or injury [1]. More than 50% of intensive care unit (ICU) patients develop AKI (defined by Kidney Disease: Improving Global Outcomes [KDIGO] criteria), with ≥30% of these patients reaching the more severe KDIGO stages (stage 2 and 3) [2, 3]. Multiple studies have identified AKI as an important risk factor for high morbidity and mortality. Furthermore, it significantly increases the need for renal replacement therapy (RRT), hospital costs, and leads to end-stage renal disease (ESRD) or chronic kidney disease (CKD) [4, 5]. Despite increasing attention in recent years, little improvement in outcomes of AKI has occurred. There are two major challenges, the difficulty to detect AKI early and the poor understanding of its pathogenesis, have hampered the progress in AKI research and clinical management [1]. Current definitions of AKI including the risk, injury, failure, loss of function, ESRD, the Acute Kidney Injury

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Network, and the KDIGO criteria all rely on changes in either the levels of serum creatinine (sCr) and/or urinary output [6–9]. sCr and oliguria are neither sensitive nor specific. Because both of them are markers of kidney function not of kidney injury or stress, and are easily influenced by many factors (including sex, muscle mass, medications or volume status), so using them may delay the diagnosis of AKI [1, 10]. Over the past decade, there have been numerous studies dedicated to discovering novel makers for an early detection of AKI in order to reverse the adverse outcomes of AKI. These biomarkers include neutrophil gelatinase-associated lipocalin (NGAL), interleukin-18 (IL-18), liver-type fatty acid binding protein (L-FABP), kidney injury molecule-1 (KIM-1), tissue inhibitor of metalloproteinases-2 (TIMP-2), and insulin-like growth factor-binding protein 7 (IGFBP7) [11–14]. Among all these markers, [TIMP-2] · [IGFBP7] shows the best accuracy and stability, even in patients with chronic conditions such as diabetes mellitus, congestive heart failure and CKD [15]. In 2014, the Food and Drug Administration (FDA) approved the test “NephroCheck®” (Astute Medical, San Diego, CA, USA) ([TIMP-2] · [IGFBP7], united in $[\text{ng/mL}]^2/1000$) to be used in ICU patients to predict the risk of developing moderate to severe AKI within the prior 24 h [16]. Since then, more and more studies focused on the evaluation of the clinical application of NephroCheck®. In this review, we summarize the current research status of cell cycle arrest biomarkers (TIMP-2 and IGFBP7), and discuss the advantages and limitations of using these biomarkers.

Biological characteristics

TIMP-2 has a molecular weight of approximately 24 kDa and IGFBP7 has a molecular mass of 29 kDa [17]. Both of them are expressed and secreted by renal tubular cells, and involved in G1 cell cycle arrest during the early phases of cellular stress or injury caused by various insults (e.g. sepsis, ischemia, oxidative stress and toxins) [18].

In addition to the quiescent state (G0), the cell cycle includes four tightly controlled phases: G1, S (DNA synthesis), G2, and M (mitosis) [19]. Each phase of the cell cycle has a specific function for appropriate cell proliferation. Cells must enter and exit each phase of the cell cycle on schedule in order to divide and repair. This process is controlled by cyclins, cyclin-dependent kinases (CDK), and cyclin-dependent kinases inhibitors. If the cells stay in a phase too long or exit a phase too soon, the normal division and repair process can become maladaptive [20]. For instance, if the cells remain arrested in G1 or G2 phase instead of re-initiating the cell cycle, a senescent,

hypertrophic and fibrotic cell phenotype will present. Conversely, exiting from the cell cycle in late G1 phase may lead to cell apoptosis [21].

When exposed to cellular stress or injury, renal tubular cells may produce and release TIMP-2 and IGFBP7. TIMP-2 stimulates p27 expression and IGFBP7 directly increases the expression of p53 and p21. Then these p proteins block the effect of cyclin-dependent protein kinase complexes (CyclD-CDK4 and CyclE-CDK2) on cell cycle promotion, resulting in transient G1 cell cycle arrest, thereby providing cells an opportunity to repair DNA damage and regain function. This process happens during early cellular stress and may help cells maintain energy balance, prevent further DNA damage and division [22, 23]. But sustained cell cycle arrest will result in a senescent cell phenotype and lead to fibrosis. So cell cycle arrest is not only associated with increased risk for AKI but may also serve as a mechanistic link between AKI and CKD [19, 24] (Figure 1).

Clinical research

Before clinical application of NephroCheck®, there have been three main studies for detecting and validating the ability of [TIMP-2] · [IGFBP7] to pre-diagnose AKI. These studies are summarized below and in Table 1.

The Sapphire study was a multicenter observational study in heterogeneous critically-ill patients, which had two phases: discovery phase and validation phase. The primary endpoint was moderate-severe AKI (KDIGO stage 2–3) within 12 h of sample collection. The purpose of the discovery phase study was to identify novel potential biomarkers for AKI. In this phase, 522 adult ICU patients were enrolled and 340 biomarkers in the urine and blood were tested (including NGAL, KIM-1, IL-18, L-FABP). As shown by the results, TIMP-2 and IGFBP7 had the best performance, with an area under the receiver operating characteristic curve (AUC) of 0.80 for [TIMP-2] · [IGFBP7] (0.79 and 0.76, respectively), which was significantly superior to all previously described markers of AKI ($p < 0.002$) [23]. Subsequently, the outcome was validated in the validation phase study which included a heterogeneous sample of 728 critically-ill patients. The combined urine biomarker product of [TIMP-2] · [IGFBP7] was proved to be stable across clinical syndromes and significantly improved risk stratification [1].

The Opal study was a derivation and validation study to confirm the accuracy and clinical utility of two different cutoff values of [TIMP-2] · [IGFBP7] in 154 adult critically-ill patients from six sites in the US. In this study, the researchers set the cutoff of $0.3 (\text{ng/mL})^2/1000$ for

Figure 1: Mechanism of TIMP-2 and IGFBP7 in AKI.

Table 1: Derivation and validation studies of TIMP-2 and IGFBP7 for predicting AKI before clinical application.

Study name, number	Goal	Study population	End point	Main results
Sapphire study				
Discovery phase (522)	Identify novel biomarkers for detecting AKI	ICU patients >21 years old, with at least one risk factor for AKI	Moderate or severe AKI (KDIGO stage 2–3)	340 potential biomarkers were tested, TIMP-7 and IGFBP7 were the best performing markers (AUC: 0.79 and 0.76, respectively)
Validation phase (728)	Validation the performance of TIMP-2 and IGFBP7 for detecting AKI			14% reached primary end point; risk of AKI was significantly elevated with [TIMP-2] · [IGFBP7] >0.3
Opal study (154)	Derivated and validate two different cutoff values of [TIMP-2] · [IGFBP7]			18% reached primary end point; AUC was 0.79. For 0.3 cutoff, sensitivity was 89%, and NPV was 97%. For 2.0 cutoff, specificity was 95% and PPV was 49%.
Topaz study (420)	Validate the performance of [TIMP-2] · [IGFBP7] with clinical adjudication		Moderate or severe AKI (judged by three nephrologist who blinded to the results)	17.4% reached primary end point; AUC was 0.82. For 0.3 cutoff, sensitivity was 92% and specificity was 46%; For 2.0 cutoff, sensitivity was 46% and specificity was 95%. [TIMP-2] · [IGFBP7] remained significant when combined with clinical model.

TIMP-2, tissue inhibitor of metalloproteinases-2; IGFBP7, insulin-like growth factor-binding protein 7; AKI, acute kidney injury; ICU, intensive care unit; AUC, area under curve; NPV, negative predictive value; PPV, positive predictive value.

high sensitivity/high negative predictive value (NPV) and 2.0 (ng/mL)²/1000 for high specificity/high positive predictive value (PPV). The results of the Opal study replicated those of the Sapphire study where the sensitivity at the 0.3 cutoff was 89%, and NPV was 97%. For 2.0 cutoff, specificity was 95% and PPV was 49% [25].

The Topaz study enrolled 420 heterogeneous critically-ill patients in order to prospectively validate the lower (0.3) cutoff value for risk assessment of AKI. In this study, the endpoint was determined independently by clinical adjudication by three nephrologists who were blinded to the results of the test. [TIMP-2] · [IGFBP7] significantly improved risk assessment, with a seven-fold increase in risk for patients with [TIMP-2] · [IGFBP7] value >0.3 compared with those ≤0.3 [26].

Thus, urinary [TIMP-2] · [IGFBP7] has now been shown to provide early detection and risk stratification for imminent AKI in over 1800 heterogeneous critically-ill patients. Since the publication of the Sapphire, Opal, and Topaz studies, NephroCheck® was approved by the FDA in 2014 to help determine whether critically-ill patients are at risk of development of moderate to severe AKI, both in the United States and Europe. From then on, more and more studies have been dedicated to clarifying the validity of [TIMP-2] · [IGFBP7] for predicting AKI in ICU patients. Some of the results are positive, such as those reported by Di Leo et al. demonstrating that [TIMP-2] · [IGFBP7] at ICU admission has a good performance in predicting AKI [27]. In contrast, some results are negative, for example, Bell et al. found that [TIMP-2] · [IGFBP7] did not predict AKI within 12–48 h and were significantly affected by comorbidities (e.g. diabetes) [28]. Additionally, some studies aimed to identify the utility of [TIMP-2] · [IGFBP7] in multiple clinical settings.

Research of [TIMP-2] · [IGFBP7] in AKI associated with different etiologies

AKI is a multi-etiological disease, many studies have tried to clarify the practicality of urinary [TIMP-2] · [IGFBP7] in AKI of different etiologies. The major and recent studies in this regard are listed in Table 2.

Cardiac surgery

Patients undergoing cardiac surgery are at high risk for AKI. A recent meta-analysis estimated the global

incidence of AKI following cardiac surgery in adults to be approximately 22% [49]. Most current clinical studies on [TIMP-2] · [IGFBP7] focus on cardiac surgery-associated AKI. In Meersch et al.'s study, [TIMP-2] · [IGFBP7] had an AUC of 0.84 for predicting AKI stage 2–3 after cardiac surgery, whereas, sensitivity and specificity were 0.92 and 0.81, respectively, for a cutoff value of 0.50. Additionally, they demonstrated that decline in urinary [TIMP-2] · [IGFBP7] values was an accurate predictor for renal recovery [29]. At the same time, previously published cutoff points of 0.3 and 2.0 could not be confirmed in Wetz et al.'s study cohort. In contrast, they found a cutoff point of 1.1 with an AUC of 0.71 (sensitivity was 0.47, specificity was 0.96) [30]. At the same time, Pilarczyk et al. found an AUC of 0.861 (sensitivity 0.83, specificity 0.67) for predicting AKI stage 2–3 4 h after surgery at cut-off 0.15 [31]. Dusse et al. found an AUC of 0.97 (sensitivity 1.00, specificity 0.90) for predicting AKI stage 2–3 on day 1 after transcatheter aortic valve implantation surgery at cut-off 1.03 [32]. Wang et al. validated the performance of [TIMP-2] · [IGFBP7] in a Chinese population of cardiac surgery patients. They concluded that [TIMP-2] · [IGFBP7] 4 h after postoperative ICU admission identifies patients at risk of developing AKI, the AUC was 0.83 [33]. Oezkur et al. investigated the association of [TIMP-2] · [IGFBP7] at various time points with the incidence of AKI in a prospective study enrolling 150 cardiac surgery patients. They demonstrated that measurement of [TIMP-2] · [IGFBP7] at ICU admission directly after surgery is a strong and accurate predictor of AKI within 48 h after surgery [34]. Levante et al. [35] and Mayer et al. [36] confirmed the ability of [TIMP-2] · [IGFBP7] for predicting AKI in their studies. In contrast, a recent study by Zaouter et al. failed to demonstrate the ability of [TIMP-2] · [IGFBP7] for predicting cardiac surgery-associated AKI occurring in the first post-operative week within the first 24 postoperative hours [37]. Therefore, although many studies have investigated the ability of [TIMP-2] · [IGFBP7] to predict AKI after cardiac surgery, the results (cutoff, test time, AUC) are inconsistent and further large-scale studies are needed.

Major surgery

AKI also commonly complicates high-risk non-cardiac surgery which has received much less attention than cardiac surgery [50]. As demonstrated in Gocze et al. and Gunnerson et al.'s study, the [TIMP-2] · [IGFBP7] was a strong predictor of AKI and significantly improved the risk assessment. The AUC for the risk of AKI was 0.85 and 0.84 in each study. In Gocze et al.'s study, they also conducted

Table 2 (continued)

The cause of AKI	Study	Patient population	AKI diagnostic criteria	AKI threshold	No. of patients enrolled/no. of patients developed AKI	[TIMP-2] · [IGFBP7] detection time	AUC	Cut off	Sensitivity	Specificity
Toxic renal disease	Schanz et al. [47] (2017; Germany)	Patients with malignant neoplastic disease, therapy with cisplatin or carboplatin	KDIGO	AKI stage ≥ 1 within 72 h after the administration of chemotherapy	58/4	Within 12 h after chemotherapy administration	0.92	0.3	0.50	0.87
	Toprak et al. [48] (2017; Turkey)	Patients with lung cancer with cisplatin containing chemotherapy	AKIN	AKI within 48 h	45/13	24 h after cisplatin administration	0.46	NR	NR	NR

ADHF, acute decompensated heart failure; AKI, acute kidney injury; AKIN, Acute Kidney Injury Network; AUC, area under the receiver operating characteristic curve; CABG, coronary artery bypass surgery; CPB, cardiopulmonary bypass; DGF, delayed graft function; ICU, intensive care unit; KDIGO, Kidney Disease: Improving Global Outcomes; KT, kidney transplantation; NR, not report; OHCA, out-of-hospital cardiac arrest; TAVI, transcatheter aortic valve implantation; TIMP-2, tissue inhibitor of metalloproteinases-2; IGFBP7, insulin-like growth factor-binding protein 7.

an AUC for early use of RRT (0.83) and for 28-day mortality (0.77) [38, 39]. So using [TIMP-2] · [IGFBP7] may not only predict AKI, but can also guide the early initiation of kidney protection treatment and predict AKI prognosis.

Kidney transplantation

Pianta et al. assessed the utility of [TIMP-2] · [IGFBP7] and five inflammatory markers to predict delayed graft function (DGF) following deceased-donor kidney transplantation in 56 recipients. Only TIMP-2 and vascular endothelial growth factor-A, not [TIMP-2] · [IGFBP7], significantly enhanced the DGF prediction at 4 and 12 h [40]. In contrast, Yang et al. indicated that [TIMP-2] · [IGFBP7] test immediately after transplantation could be an early, predictive biomarker of DGF in kidney transplantation, with an AUC of 0.867 (sensitivity 0.86, specificity 0.71) for a cutoff value of 1.39 [41].

Decompensated heart failure

Acute decompensated heart failure (ADHF) is another disease associated with a high risk of AKI, but the research in this area is limited. In Schanz et al.'s study, they examined the predictive ability of urinary [TIMP-2] · [IGFBP7] for development of AKI stage 2 or 3 within 24 h of sample collection. Of the ADHF patients 27.5% developed AKI stage 2–3 within 7 days. Urinary [TIMP-2] · [IGFBP7] discriminated AKI stage 2–3 over the first day with an AUC of 0.84, sensitivity was 86% at the 0.3 cutoff and specificity was 95% at the 2.0 cutoff. They concluded that in patients with ADHF, urinary [TIMP-2] · [IGFBP7] is associated with moderate to severe AKI and related to increased mortality [42].

Cardiac arrest

Patients after cardiac arrest are predisposed to development of multiple organ failure, especially AKI, due to ischemia-reperfusion injury. [TIMP-2] · [IGFBP7] levels only predicted AKI in urine samples collected at admission, but, was not significantly associated with the development of AKI at day 3 as confirmed by Beitland et al.'s study [43]. In a recent research study, Adler et al. found that urinary [TIMP-2] · [IGFBP7] reliably predicts AKI in high-risk patients only 3 h after determination of cardiac arrest with a cut-off at 0.24 [44]. Research in this area is still very limited. Further larger-sample studies are needed

to confirm which biomarker has better clinical utility of predicting AKI.

Sepsis

Sepsis is a major cause for AKI. In fact, almost half of AKI is caused by sepsis [51, 52]. The [TIMP-2] · [IGFBP7] test provides accurate prediction of AKI in septic patients and the test performance is not affected by non-renal organ dysfunction, as was confirmed in Honore et al.'s study [45]. Cuartero et al. presented a prospective, observational study including 98 ICU patients to examine the role of [TIMP-2] · [IGFBP7] in septic AKI and non-septic AKI. The AUC to predict AKI was 0.798 (sensitivity 73.5%, specificity 71.4%). [TIMP-2] · [IGFBP7] was found to be an early predictor of AKI in ICU patients regardless of sepsis. Moreover, [TIMP-2] · [IGFBP7] values <0.8 (ng/mL)²/1000 ruled out the need for RRT [46]. This study showed that [TIMP-2] · [IGFBP7] is associated with AKI, but is not specific for sepsis.

Toxic renal disease

Platinum-based chemotherapy (PBC) is broadly used potent antineoplastic treatments, with potential nephrotoxicity, especially of cisplatin. Schanz et al. conducted a clinical observational study enrolling 58 patients with malignant neoplastic disease, four (12.5%) patients developed AKI within 72 h. In their study, urinary [TIMP-2] · [IGFBP7] values after the administration of PBC were significantly higher in patients with AKI than those without AKI. The AUC was 0.92. At the cutoff of 0.3 for [TIMP-2] · [IGFBP7], sensitivity was 50%, specificity was 87%, NPV was 95% and PPV was 25% for the prediction of AKI within 72 h. So they concluded that urinary [TIMP-2] · [IGFBP7] measured after PBC may be a useful tool for early identification of patients at risk of developing platinum-induced AKI [47]. In contrast, in Toprak et al.'s study, those findings were not replicated [48]. Overall, these two studies had small sample size. Larger studies are required to determine whether [TIMP-2] · [IGFBP7] can predict cisplatin-related AKI.

Future directions

To date, the published literature about [TIMP-2] · [IGFBP7] mainly focuses on the application of [TIMP-2] · [IGFBP7] in adult ICU patients who develop ischemic or

nephrotoxic-AKI as already described. Pajenda et al. tested [TIMP-2] · [IGFBP7] in 69 patients with different settings of AKI. They indicated that in patients with ischemic reperfusion and toxic injury, the values of [TIMP-2] · [IGFBP7] rise and decline rapidly, even before polyuria becomes evident. Furthermore, they confirmed that when [TIMP-2] · [IGFBP7] value remains below 8, the concomitant rise in sCr and the stage of AKI appear to be reversible. Conversely, [TIMP-2] · [IGFBP7] value of more than 18 that remained elevated for a long time indicated that the kidney function is unrecoverable and patients would require RRT permanently [53]. Whether [TIMP-2] · [IGFBP7] use can be expanded in other contexts or patient populations, such as pediatric patients has not been confirmed.

The performance data of [TIMP-2] · [IGFBP7] in different patient populations outside the ICU or perioperative setting is still lacking [13]. Some studies showed that [TIMP-2] · [IGFBP7] provides independent prediction of AKI in emergency department patients as well [54, 55]. As far as its use in the pediatric population, one study looking at [TIMP-2] · [IGFBP7] level following cardiac surgery implied that [TIMP-2] · [IGFBP7] alone is not suitable for predicting AKI in this patient population [56]. Another study demonstrated that [TIMP-2] · [IGFBP7] can be used in infants to predict AKI following cardiopulmonary bypass [57]. Meanwhile, more studies are needed to confirm the clinical utility of [TIMP-2] · [IGFBP7] in other causes of AKI (e.g. acute interstitial AKI, post-renal AKI).

Recognizing the major risks associated with AKI and in an attempt to potentially reverse its adverse outcomes, the FDA has taken an important step to provide us with a new tool as an early alert of which patients are at imminent risk [58]. However, because AKI is often multifactorial, it seems unlikely that a single AKI biomarker would achieve troponin-like diagnostic accuracy. So like all diagnostic tests, NephroCheck® is not a standalone test, it needs to be combined with clinical judgment (such as urine sediment score) in order to detect AKI with higher sensitivity and specificity [59]. Therefore, in future studies, how to combine NephroCheck® test with other clinical examinations or symptoms to improve diagnostic accuracy is an important research direction.

TIMP-2 and IGFBP7 do not appear to persist in the urine for long after AKI, meanwhile, the combination of them is more sensitive to transient AKI with a higher delta NephroCheck® score comparing to persistent AKI [60]. Furthermore, the concentration of [TIMP-2] · [IGFBP7] does not depend on the expression of *TIMP-2* and the *IGFBP7* gene in cells of the urinary sediment. In addition, there is no correlation between [TIMP-2] · [IGFBP7] with sCr, BUN or eGFR [61]. Therefore, NephroCheck® may be normal in patients

who have already manifested AKI by functional criteria (e.g. sCr). AKI complicating critical illness is highly heterogeneous in severity, etiology and timing, so defining the appropriate timing and frequency of biomarker measurement and interpreting these results in individual patients is extremely difficult [62]. However, it is very meaningful to verify the best time and frequency of NephroCheck® test, for example, it can save unnecessary testing costs.

Some researchers suggest that [TIMP-2] · [IGFBP7] for the prediction of AKI might be most applicable in patients at high risk of AKI, and less precise in those at lower risk [63]. Among patients with AKI risk, applying high-sensitivity threshold could focus patient care on strategies of intensified monitoring prior to any increase in sCr. Furthermore, combining AKI diagnostic criteria (such as KDIGO criteria) with NephroCheck® test results may enable us to predict the development trend of AKI, recovery or progress to the next phase of AKI. In this way, NephroCheck® could be utilized and integrated with additional clinical information to help inform more complex and invasive management decisions, such as prediction of the need for RRT, and deciding when to start or stop RRT [64]. Meanwhile, the cost benefit of [TIMP-2] · [IGFBP7] test is unknown. We need more clinical trials and data to support the hypothesis that early recognition of kidney injury with [TIMP-2] · [IGFBP7] will prevent the progression of AKI or be associated with a cost benefit to the patient or institution, avoiding unnecessary and expensive diagnostic and therapeutic evaluations.

Furthermore, measurement of [TIMP-2] · [IGFBP7] not only provides information for predicting and diagnosing AKI, but also has the potential to help to identify patients who are at the highest risk of developing adverse outcomes [65–67]. Further studies to evaluate the predictive value of [TIMP-2] · [IGFBP7] for ICU stay, mortality and progression to ESRD, are needed. Moreover, the current studies looking into the mechanism by which the test works is still in the hypothesis phase, additional studies using conditional knockouts and pharmacologic inhibitors of TIMP-2 and IGFBP7 are needed to better define its mechanistic role in renal injury [68]. As a result, cell cycle regulation may become a potential new target for the prevention and treatment of AKI. We may even be able to prevent CKD progression by regulating cell cycle arrest [69].

Conclusions

As a novel biomarker for predicting AKI, [TIMP-2] · [IGFBP7] has already proved its validity and accuracy in multiple

studies in many fields and has been applied by the FDA for clinical application. Further studies regarding the application value of [TIMP-2] · [IGFBP7] in different clinical contexts, different patient populations, different disease spectrum, are needed. It is important to translate this advancement in AKI biomarkers to meaningful improvement in clinical care and treatment strategy, in order to achieve better outcomes.

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